THE UNIVERSITY OF ALBERTA

UTILIZATION OF UREA-NITROGEN BY EARLY-WEANED HOLSTEIN CALVES

by

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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies for acceptance, a thesis entitled, "Utilization of Urea-Nitrogen by Early-Weaned Holstein Calves" submitted by William Kiiza Ndyanabo, B.Sc. (Agr.), in partial fulfilment of the requirements for the degree of Master of Science.

Abstract

Studies were conducted to compare calf meals containing 0.50 and 0.75 percent urea with a calf meal containing soybean meal. Each calf meal was fed to a group of four heifer and four bull Holstein-Friesian calves.

The calves received colostrum for the first three days of life after which they were fed milk replacer until weaned at 28 days of age. The calf meals were fed free-choice commencing when the calves were 10 days of age and until the studies were terminated when the calves were approximately 90 days of age. Feed consumption and growth rate were measured during the entire experimental period.

Metabolism studies were conducted with two bull calves from each treatment commencing when the calves were 72 to 76 days of age. Apparent digestibility and nitrogen retention were determined. Half carcasses of these two bull calves from each treatment were dissected, and carcass composition was determined. All calves were bled at the age of 80 to 84 days and blood urea concentrations were measured.

Calves fed the ration containing 0.75 percent urea consumed more average feed, gained more average weight daily and consumed more feed per unit weight gain than calves fed the ration containing soybean meal. Calves fed 0.50 percent urea ration had the poorest performance in weight gain and feed intake, but the difference was small. Urea did not appear to reduce acceptability of the calf meals in this experiment.

Apparent digestion coefficient of dry matter was significantly higher ($P < 0.05$) in the ration with soybean meal than in the ration with 0.50 percent urea. Apparent digestion coefficient of gross

energy was significantly higher ($P < 0.05$) in the ration containing soybean meal than in the rations containing urea. The apparent digestion coefficient of nitrogen was significantly higher ($P < 0.01$) in the ration containing soybean meal than in the ration containing 0.50 percent urea; and significantly higher ($P < 0.05$) than in the ration containing 0.75 percent urea.

Average nitrogen retention was similar for five of the six bull calves used in the metabolism studies. One calf fed the ration containing 0.50 percent urea had comparatively low nitrogen retention, which resulted in a low average value for the calves in this treatment. Some metabolic disturbance might have been responsible for the low dry matter consumption and nitrogen retention by this calf.

There were no significant differences in blood urea concentrations among the calves in all treatments, suggesting that ammonia from urea nitrogen was used for microbial protein synthesis and was not absorbed directly into the blood stream in excessive quantities.

Carcass composition of the six bull calves in all treatments was similar, suggesting that urea nitrogen was used as effectively as soybean meal nitrogen for muscle development.

It was concluded that nitrogen from urea was used for growth of the calves almost as effectively as nitrogen from soybean meal. Therefore, urea at moderate levels in calf meals appeared to be a satisfactory source of supplemental nitrogen for early-weaned calves.

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Introduction

It is estimated that 50 percent of the human population of the world is suffering from a shortage of dietary protein. The demand for protein by man, and the need for more available sources of protein for feeding other monogastric animals have been responsible for extensive investigations of alternate sources of dietary nitrogen. Urea, the most extensively fed source of non-protein nitrogen has proven of value in ruminant feeding because ruminal microflora are capable of converting its nitrogen to a nutritionally useful form.

It is well established that in mature ruminants urea is rapidly hydrolysed in the rumen to carbon dioxide and ammonia by the urease activity of the rumen microorganisms. The rapidly-released ammonia from urea hydrolysis is subsequently assimilated by the rumen bacteria in the synthesis of cell constituents. However, the rate of urea hydrolysis in the rumen is greater than the rate of fixation of liberated ammonia into cellular protein. This results in ammonia passing out of the rumen and, therefore, becoming of little benefit to the animal. Ammonia passing out of the rumen is either absorbed through the ruminal wall into the portal blood stream or passed into the lower portions of the gut. This ammonia is not entirely lost as part of it may be returned to the rumen as urea during urea recycling. In addition, under unfavourable conditions, the concentration of ammonia in the rumen may reach a level at which the rate of absorption may be greater than that at which the liver can convert it to urea; ammonia then spills over into the peripheral blood and when it reaches certain levels in blood the animal shows signs of toxicity.

In calves, the feeding of urea has not been recommended as the rumen in young calves is not sufficiently developed to permit the utilization of non-protein nitrogen. The practical recommendation has been to feed urea when the animals are four months or older. However, more recent work has shown that calves at two months of age are able to utilize some urea.

This experiment was designed to study urea utilization by early-weaned dairy calves which were weaned directly to urea-containing concentrate rations. The effects of the rations were measured on feed intake, growth rate, digestibility, blood urea concentrations and carcass composition in calves fed concentrate rations containing soybean meal or urea.

Review of Literature

Nitrogen Metabolism in the Ruminant

The feed ingested by the ruminant animal undergoes microbial fermentation in the rumen to produce the required nutrients in the form of end-products of such bacterial action. In addition to protein and non-protein nitrogen (NPN) in the diet, nitrogen enters the rumen as urea, which is contained in the alkaline saliva of the animal, while some of it passes from the bloodstream through the rumen wall (Tillman and Sidhu, 1969). These sources of nitrogen are acted on by microbes in the rumen and both dietary and endogenous urea are rapidly broken down into ammonia and carbon dioxide by bacterial urease (Pearson and Smith, 1943; Streeter _et al_. 1969). Ammonia is, in turn, used to form microbial protein for bacterial tissue growth (Annison and Lewis, 1959).

Nitrogen leaves the rumen mainly as microbial protein and ammonia, and as intact protein in the undigested feed (Figure 1). The microbial protein is digested in the stomach and intestines (Hume _et al_. 1970) to the constituent amino acids which are absorbed, and subsequently either used for animal protein synthesis or broken down to yield energy. With the energy yielding process there is a corresponding release of ammonia which is absorbed across the rumen wall and carried via the portal circulation to the liver, where it is converted to urea (Tillman and Sidhu, 1969). Urea in the blood originating from amino acid breakdown, or from ammonia absorption, can be excreted in the urine or reabsorbed into the rumen (Milligan, 1967b). Urea that is reabsorbed into the rumen is rapidly converted

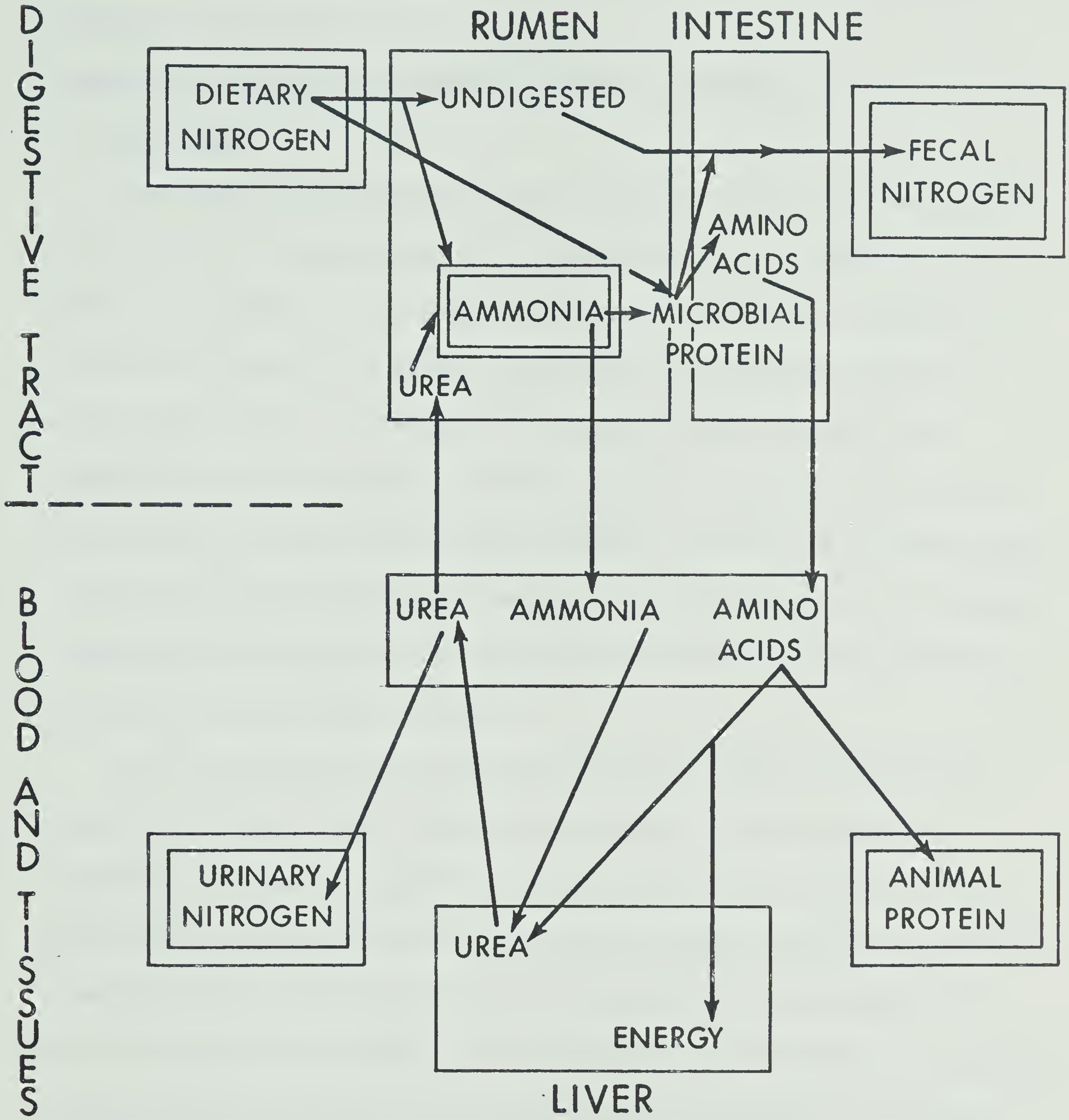

Figure 1. Nitrogen conversions in the ruminant (Milligan, 1967b).

to ammonia which may be used to maintain an active microbial population, thus completing the "Nitrogen Cycle" (Moir and Harris, 1962). The pathways of nitrogen metabolism in the ruminant are illustrated schematically (Figure 1).

Non-protein Nitrogen as a Source of Dietary Nitrogen

1. Historical

The unique role played by the microorganisms of the rumen in the nutrition of the host animal was recognized for the first time by German scientists in 1891. They outlined the idea that NPN might be converted to protein by the microorganisms of the rumen and that this protein might be subsequently digested and utilized by the ruminating animal (McNaught and Smith, 1947). Because of the importance of such a process in feeding practice, the ability of these rumen organisms to utilize NPN continued to be explored during the following period, and the theories were vigorously contested largely by German research workers (Stangel, 1967).

In 1911, Armsby of the United States Department of Agriculture (cited by Stangel, 1967) critically reviewed the first series of investigations and concluded that when the level of protein in the diet was low and other conditions were favourable, NPN could serve as a partial substitute for protein for maintenance, and possibly for milk production and growth. He also noted that non-protein nitrogenous substances were inferior in nutritive value to protein of equivalent nitrogen content. He attributed this observation to the limited ability of the rumen microorganisms to form protein rather than to inferiority of the protein so formed.

Contrary to the views of Armsby, Krebs in 1937 (cited by Hart et al., 1939) published a critical review of the literature on the subject, showing his reluctance to believe that any considerable protein synthesis occurred, or that if it did occur, the bacterial protein produced was not of any appreciable value to the host animal. However, Reid (1953) quoted Krebs admitting that the evidence indicated some protein-sparing effect of substances like urea and glycine, but he attributed the effect to neutralization of the organic acids formed in the rumen.

Since the Krebs' review in 1937, a lot of work on nitrogen balance (Harris and Mitchell, 1941a and 1941b; Loosli and McCay, 1943; McLaren et al., 1961), growth (Bartlett and Cotton, 1938; Harris et al., 1943; Hart et al., 1939; Work and Henke, 1939) and milk yield (Oltjen and Bond, 1967; Ryś, 1967) has furnished convincing evidence, in agreement with Armsby's views, that low protein rations inadequate for growth in calves, and inadequate for maintenance and growth in sheep are efficiently supplemented by urea. The work done, therefore, left no doubt that urea was converted to protein.

The fact that other sources of NPN besides urea were utilized by ruminants had been reported by European investigators (Bartlett and Cotton, 1938). However, it did not become clear that these sources were indeed effectively utilized until Hart et al. (1939) conducted an experiment from which they concluded that ammonium bicarbonate was a good source of nitrogen, as was urea when used to supplement low protein rations. This observation led to the belief that other sources of NPN might be used by rumen bacteria to produce protein. In the same experiment, Hart et al. (1939) found that the composition

of body protein of the animals studied was not affected by the source of nitrogen consumed. This observation was interpreted to mean that ruminant animals absorbed protein in the form of microbial protein which was not the same as dietary protein. This conclusion agreed with current views held about nitrogen metabolism in ruminants (Annison and Lewis, 1959; McDonald and Hall, 1957).

2. Urea Nitrogen Compared to Protein Nitrogen for Mature Ruminants

One of the most conclusive of the earlier experiments comparing protein and NPN utilization in mature ruminant animals was reported by Harris and Mitchell (1941a). They used 8 sheep aged 15 to 18 months and weighing 27 to 40 kilograms. After determining the endogenous urinary nitrogen and the fecal metabolic nitrogen, casein or urea was given for periods ranging from 6 to 34 days. The results showed that it required 202 milligrams urea nitrogen per kilogram body weight to obtain nitrogen equilibrium, whereas 161 milligrams casein nitrogen was required in similar circumstances. Therefore, the efficiency of urea nitrogen utilization was approximately 80 percent of that of casein. At nitrogen equilibrium, the biological value of the urea nitrogen was 62 and the casein nitrogen 79, but, as would be expected, the biological values decreased as the amounts of urea and casein ingested were increased.

More recently, Chalupa (1968), in a review of the literature, pointed out that nitrogen retention on diets containing protein was usually superior to that on diets containing NPN. The superior nitrogen retention on diets containing protein compared with diets containing NPN could be due to two reasons (Hume, 1970): Firstly, some undegraded dietary protein, in addition to microbial protein,

passes to the abomasum when the solubility of the dietary protein is low; and secondly, more microbial protein is formed and leaves the rumen when protein is given.

Freitag _et al._ (1968) carried out an experiment using two pairs of Angus twin steers, weighing approximately 325 kilograms, to compare nitrogen balance for steers fed either diets supplemented with urea and soybean meal or urea alone. Their results did not show the expected decrease in nitrogen balance due to feeding the urea diet compared to the diet supplemented with soybean meal and urea.

Studies with mature lactating cows were carried out by Rupel _et al._ (1943). They replaced linseed meal with urea in a concentrate mixture containing 18 percent protein and the result was a non-significant increase in milk production by the cows fed linseed meal as compared with those fed urea. Later on they added molasses to the grain and urea combination, but there was no beneficial effect. The workers concluded with a recommendation that urea should not be fed in excess of 1 percent of the dry matter in the ration or 3 percent of the concentrate mixture.

3. Urea Nitrogen Compared to Protein Nitrogen for Calves and Other Growing Ruminants

Most studies have shown that urea nitrogen is somewhat inferior to natural protein for growing animals. Bartlett and Cotton (1938) fed rations that contained normal protein, low protein and low protein plus 0.127 pounds of urea daily to 21 dairy heifers, ranging in age from 7 to 17 months for 142 days. The results showed significant differences between the low protein group and the urea group, but

showed no significant differences between urea-fed and normal protein groups. Similar results were obtained by Work and Henke (1939).

In later work with lambs, Johnson _et al_. (1942) compared the utilization of urea nitrogen with that of soybean meal and casein when those substances were given as supplements to a basal diet low in protein. When the crude protein content of the basal ration was raised from 6 to 10 or 12 percent by adding urea, so that urea supplied from 40 to 50 percent of the ingested nitrogen, a marked improvement in nitrogen retention occurred. By adding true protein to the ration, the amount of nitrogen retained was increased still further, but not when additional amounts of urea were added. These workers expressed the opinion that the nitrogen of urea was not changed to protein rapidly enough to meet the requirements of growing lambs.

In an experiment with calves, Harris _et al_. (1943) found the daily nitrogen balance was -9 grams on a basal diet containing 7.4 percent crude protein as compared with +1 gram when the basal diet was supplemented with 6 percent of urea, and 21 grams when it was supplemented with soybean meal. The biological value of urea nitrogen was estimated to be 34 as compared to 60 for soybean meal nitrogen. It, therefore, became obvious from the results of this work that urea nitrogen was markedly inferior to soybean meal protein, but the levels of urea fed in this experiment were too high to be efficiently utilized.

Loosli and McCay (1943), experimenting with calves aged 2 months, fed a ration in which 73 percent of the nitrogen was from urea and had a biological value of 52. They reported satisfactory gains in weight and height. The apparent digestibility of nitrogen in the ration was

about 80 percent, and, while calves on the basal ration were in negative nitrogen balance, calves fed the urea ration stored 24 to 36 percent of the nitrogen in the diet.

Brown et al. (1956) conducted studies to determine the usefulness of urea as a source of nitrogen for young dairy calves from 2 to 86 days of age raised on a limited milk-hay-starter system. They found that 6-week-old Jersey calves gained as rapidly and efficiently when urea supplied 54.2 percent of the total dietary nitrogen (15.1 percent crude protein) as when linseed meal was the primary source of nitrogen (15.2 percent crude protein). Although the calves were not early-weaned, their growth rate up to 86 days of age was 0.52 kilograms per day, which agreed with results reported from other experiments with early-weaned calves (Kay et al., 1967; Nelson et al., 1966).

Recently, Nelson et al. (1966) studied the effects of protein quality in starter rations on rate of gain, using 64 Holstein calves, from 4 to 84 days of age. Four calf starters which contained different nitrogen supplements were fed: Soybean meal (SBM), soybean meal plus α-hydroxymethionine analog (SBM - HMA), soybean meal plus urea (SBM - urea) and urea. The results showed that SBM was a superior protein supplement to all other supplements or combinations of supplements used in the experiment. Feed consumption was not significantly different in groups fed SBM and urea, but calves fed soybean meal consumed significantly more feed than those fed SBM - urea. Feed utilization was poorest for the group fed urea and best for the group fed SBM - HMA.

Similar results were reported by Miron et al. (1968), who fed Holstein calves rations supplemented with soybean meal or urea as

sources of nitrogen, and branched-chain volatile fatty acids. Their results showed that the substitution of soybean meal by urea in calf starters resulted in slower, but satisfactory gains. They pointed out that the use of urea in calf starters may be desirable under conditions when maximum gains are not necessary. Kay et al. (1967) had earlier reported essentially the same results in their experiment with Friesian bull calves, which they fed rations containing fish meal or urea as primary sources of nitrogen in the rations. Stobo et al. (1967) reported similar observations when they fed Ayrshire and Shorthorn bull calves urea- or fish meal-supplemented diets to study the ability of the calf weaned at 5 weeks to utilize urea added to a low protein concentrate. The use of purified diets to study nitrogen metabolism and utilization in ruminants (Oltjen and Bond, 1967; Oltjen et al., 1969) have resulted in calves fed purified diets with urea having growth rate and nitrogen retention as good as calves fed purified diets containing isolated soy protein.

Potter et al. (1969) carried out studies on the nature and quantity of nitrogen compounds reaching the abomasum of steers fed soybean meal or urea in a finishing ration. Their results showed that significantly more total nitrogen was recovered in the abomasum of steers fed soybean meal than in steers fed urea. In addition it was observed that more of the nitrogen in the abomasum was in the form of protein and less as free amino and purine-pyrimidine nitrogen, when soybean meal was fed than when urea was fed.

Factors Affecting Urea Nitrogen Utilization

1. Urea Toxicity

The question of whether feeds containing large amounts of urea

could cause injury to cattle and sheep was often raised by livestock producers. This problem was first given serious attention during the investigation by Hart et al. (1939). The authors reported pathological changes in the kidneys and livers of dairy heifers on high urea rations for extended periods. The report caused some apprehension concerning the use of urea as a protein supplement in ruminant rations, but there remained some confusion regarding a safe level because other experiments, where higher levels of urea were used, did not reveal liver or kidney damage (Harris and Mitchell, 1941b; Work and Henke, 1939; Work et al., 1943). At that time the level of urea recommended and used in practical rations was considerably under the level at which acute or chronic toxicity was expected. However, the level of urea in low-protein rations, such as corn cob meal, was approaching that point (Stangel, 1967). Continued investigations (Briggs et al., 1947; Woodward and Shepherd, 1944) failed to reveal any evidence of damage to kidneys or other visceral organs.

Gullup et al. (1953) made excellent descriptions of the urea toxicity symptoms and described effective antidotes. In addition they confirmed that urea toxicity was likely to occur under certain unusual feeding conditions, such as rapid consumption of urea-containing feeds by starved or fasted animals, rapid consumption of urea-containing feed by animals not previously fed urea-containing feeds, low protein rations, or rapid consumption by aggressive animals.

It is now known that when urea is fed in substantial quantities, the rate of ammonia production from its degradation can greatly exceed the rate of microbial incorportion of ammonia. This leads to a build-up of rumen ammonia and direct absorption of ammonia into the

blood. It must then be reconverted to urea after passing into the liver (Lowe, 1967). Dinning et al. (1948) reported that urea in amounts exceeding 100 grams, when administered as a drench to steers, produced a rapid rise in the levels of both urea and ammonia of the systemic blood. They reported that ataxia appeared in steers when ammonia nitrogen of the systemic blood reached a level of approximately 2.5 milligrams percent. However, when given as feed mixed in a concentrate ration, urea in amounts up to 400 grams daily did not produce any detrimental effects in steers. Lewis et al. (1957), in an experiment with sheep, reported that when ammonia levels rose too high (above 60 mmoles per litre in sheep rumen contents, or as the concentration of ammonia in peripheral blood exceeded 0.6 to 0.9 mmoles per litre) symptoms of ammonia toxicity appeared through direct action of ammonia on tissues and disturbances of blood acid-base balance.

Lowe (1967) recommended that urea must be fed to ruminants either in small quantities or, when in relatively large quantities, at a controlled rate so that no massive intake of urea at any one time was possible and rumen ammonia did not reach toxic levels. In addition, urea feeding was best introduced at low levels and gradually increased to allow the liver to adjust to the increased ammonia load, and the rumen microorganisms to the metabolism of urea.

2. Effect of Carbohydrate and Energy Content of the Ration

Rumen microorganisms have a definite energy requirement and the degree to which this requirement is met influences the utilization of urea. Wegner et al. (1940) showed that the addition of readily available carbohydrates such as molasses, dextrose or starch

stimulated the disappearance of inorganic nitrogen. Mills et al. (1942) found that a high cellulose diet such as timothy hay was a poor medium for the bacterial synthesis of protein from urea. Further studies (Mills et al., 1944) showed that the addition of starch or molasses to timothy hay promoted more efficient utilization of urea. However, the addition of starch as well as molasses to the ration resulted in additional improvement. The authors concluded that urea could be utilized at a maximum rate and efficiency in the rumen only when fermentable carbohydrate (starch) was included in the ration.

Contrary views on the subject were held by other workers (Bell et al., 1951; Rupel et al., 1943; Willett et al., 1946) who found that the addition of molasses to rations already containing starch in the form of corn, barley or oats did not improve urea utilization. However, British workers (Arias et al., 1951; McNaught and Smith, 1947; Pearson and Smith, 1943; Smith and Baker, 1944) further concurred with the first group of workers who advocated the increased benefit resulting from the addition of starch, as well as molasses to the ration. They also investigated the effect of a number of other carbohydrate sources and found that none of them proved to be more effective than starch. They concluded that small amounts of readily available carbohydrate stimulated both urea utilization and cellulose digestion.

While most workers (Arias et al., 1951; Pearson and Smith, 1943) suggested that the slower disappearance of starch from the rumen was one reason for its superiority to molasses, Annison et al. (1954) suggested that the encouragement of lactobacilli was a more plausible reason. These bacteria have been shown to use ammonia even in the

presence of peptones (Stangel, 1967). Thus, rapidly fermented sugars in cane molasses were found to disappear too quickly from the rumen, and hence were inferior in promoting nitrogen utilization by steers fed rations in which urea furnished 33 percent of the nitrogen (Mills et al., 1944). On the other hand, cellulose became available too slowly to satisfy energy needs of microorganisms (Belasco, 1956; Blaxter and Wainman, 1964). A mixture of readily available and more complex, slowly available carbohydrates appeared to be satisfactory (Karr et al., 1966; Oltjen and Putnam, 1966).

The need for a general agreement as to type and quality of concentrate mixture or forage with which urea-containing mixtures should be fed to give maximum benefits has received attention in research. Some feed manufacturers believe that urea can be utilized most effectively in high quality, low fibre concentrate mixtures; whereas others use it in economy-type high fibre mixtures. Colovos et al. (1963) fed dairy heifers two types of concentrate mixtures, a low fibre mixture and a high fibre mixture, each of which contained varying levels of urea. Their results showed that without urea, the low-fibre ration was superior to the high-fibre ration in digestible energy and total digestible nutrients. The addition of 40 pounds of urea per ton made the high-fibre ration comparable to the low-fibre ration. As the urea in the ration was increased, the heat increment decreased. This suggested that the inclusion of urea in the concentrate mixture for dairy cattle fed high-fibre rations tended to increase the net energy value of the ration, apparently due to a decreased loss of energy in the heat increment.

3. Effect of Protein Content of the Ration

The main factors of ration protein that influence the degree of urea utilization in ruminants can be considered in three aspects: the protein level in the ration; the nature of food protein in the ration; and the addition of amino acids such as lysine or methionine.

The efficiency of the process whereby urea nitrogen is transformed into microbial protein is partly dependent upon the protein level of the diet. Wegner *et al*. (1940, 1941) were among the first to demonstrate that the level of protein in the ration influenced the conversion of inorganic nitrogen to protein. They also found that as the level of protein in the ration was increased, the amount and rate of conversion of urea to protein decreased. Bell *et al*. (1953) working with steers, and Harris and Mitchell (1941b), working with sheep, independently showed that urea was more efficiently digested and utilized than a whole protein supplement, when it was used in a ration containing 7 to 11 percent protein. Balch and Campling (1961) reported essentially the same results from experiments with dairy cows.

The properties of the protein in the supplementary feeding stuff are of basic importance for urea utilization. Only those proteins which undergo proteolytic reconstruction with difficulty in the rumen can bring about an apparent protein starvation necessary for proper utilization of urea (Ryś, 1967). McNaught and Smith (1947) also pointed out that an insoluble protein such as zein in the ration resulted in a small amount of ammonia being formed from the protein, thus favouring the more efficient utilization of urea.

Results from three experiments conducted by Gossett *et al*. (1962) designed to study the effect of adding lysine to high-urea fattening

rations for beef steers indicated the possibility of better urea utilization when lysine was added. Hale et al. (1959) reported a 15 percent increase in average daily gain of beef cattle by adding 10 grams of L-lysine-HCl per head daily to a ration containing very little supplemental urea. The addition of lysine was also beneficial in a pelleted lamb fattening ration containing 1 percent urea.

Experimental results obtained by adding methionine to rations containing urea have been inconsistent. Loosli and Harris (1945) and Lofgreen et al. (1947) showed a significant increase in nitrogen retention while Klosterman et al. (1951) and Noble et al. (1955) did not show any significant improvement. Barth et al. (1959) have shown that when 17 percent of the urea nitrogen was replaced by an equivalent amount of methionine, with tryptophan added at the same time, better utilization of nitrogen was obtained in sheep fed a half-synthetic diet with 87 percent of the total nitrogen enriched by urea.

4. Influence of Mineral Supplementation

Mineral supplementation of urea-containing rations has received attention from research workers, whose results have been characterized by lack of agreement. Plumlee et al. (1953) reported depressed appetite of twin beef calves fed a mineral-supplemented wintering ration of ground corn cobs. In high-urea rations, trace mineral fortification did not give evidence of improving daily gain or feed efficiency of Hereford steer calves (Gossett et al., 1962). It has been reported, however, (Ward et al., 1955) that most of the feeds fed in urea-supplemented rations contain sufficient amounts of sulphur and trace minerals, and added improvement is not observed when urea

rations are supplemented with these minerals.

Thomas et al. (1951) produced a sulphur deficiency in growing lambs fed a purified diet low in sulphur and found that urea nitrogen was not utilized in the absence of dietary sulphur; deficient lambs were consistently in negative nitrogen and sulphur balance. Elemental sulphur was successfully used as a supplement to a sulphur-deficient diet containing urea and gelatin as the protein source by Starks et al. (1953). They pointed out that sulphate might be preferable to sulphur because of its solubility.

Beeson (1965) listed factors known to be essential for optimal utilization of high-urea supplements, and the following referred specifically to mineral supplementation:

(i) Adequate levels of calcium and phosphorus. High urea rations are usually deficient in both calcium and phosphorus.

(ii) Special attention should be given to supplying the proper level of trace minerals, especially cobalt and zinc.

(iii) Sulphur may become a limiting factor for the microbial synthesis of methionine and cysteine. Experimental evidence indicates that the nitrogen:sulphur ratio should not be wider than 15:1.

(iv) A 3.5% salt to improve the palatability of high-urea supplements.

Experiments at The University of Alberta

These experiments were designed to study urea utilization in dairy calves which were early weaned to urea-containing rations. The specific traits studied were:

1. Feed intake
2. Growth (daily weight gain)
3. Digestibility and nitrogen retention
4. Blood urea nitrogen concentrations
5. Carcass composition

Experimental

Experimental Calves

Twenty-four Holstein-Friesian calves consisting of equal numbers of males and females from the herd at the Dairy Cattle Research Unit, The University of Alberta, Edmonton Research Station were used in the experiment. At birth each calf was assigned to one of three treatments, so that each treatment contained 4 heifer and 4 bull calves. The calves in each treatment were fed the experimental rations until approximately 90 days of age at which time the experiment was terminated.

Experimental Rations

Three calf meals were formulated (Table 1). The control ration fed to calves in Treatment I contained soybean meal to raise the protein level to 13.5 percent, which was considered to be close to the minimum requirement of crude protein for growth in young calves (Agricultural Research Council, 1965; Gardner, 1968; National Research Council, 1966). The ration fed to calves in Treatment II contained no soybean meal, but 0.50 percent urea was added as a source of supplemental nitrogen, to determine whether the non-protein nitrogen would be utilized for muscle

development. In the ration fed to calves in Treatment III, the urea was increased to 0.75 percent to determine whether additional non-protein nitrogen would result in faster rate of growth. Dried molasses was increased with each increase in the level of urea to minimize the possibility of urea lowering acceptability of the ration and, therefore, resulting in reduced feed intake by the calves.

Table 1

Formulation of experimental rations.

Treatment Feed ingredients (kg)	I Soybean meal	II 0.5% urea	III 0.75% urea
Dried molasses	5.00	7.50	10.00
Wheat	60.90	60.40	57.65
Soybean meal	2.50	-----	-----
Dehydrated alfalfa	5.00	5.00	5.00
Oats	25.00	25.00	25.00
Urea	-----	0.50	0.75
Limestone	0.75	0.75	0.75
Vitamin A, 10,000 IU/g Vitamin D, 35,000 IU/g Vitamin E, 20,000 IU/g	0.09	0.09	0.09
Vitamin B mix	0.20	0.20	0.20
Aureomycin, 55.125 g/kg	0.06	0.06	0.06
Cobaltized-iodized salt	0.50	0.50	0.50
TOTAL	100.00	100.00	100.00
Analysis (on air dry basis)			
Dry matter, %	88.91	88.19	88.91
Crude protein, (N X 6.25) %	13.53	14.13	14.39
Gross energy, Mcal/kg	4.09	4.09	4.04

Feeding and Management

All calves received colostrum and were offered 2.5 kg of whole milk at each of two daily feedings until 5 days of age. From 6 to 28 days of age, the calves were fed a commercial milk replacer containing

20 percent fat and 21 percent crude protein offered at a maximum level of 225 grams at each of two daily feedings until the calves were 21 days of age. From 22 to 28 days of age, the calves were fed 150 grams of milk replacer per day, and were weaned at 28 days of age. The milk replacer was mixed with warm water in the proportions of 1:7 (w/w).

The calf meals were fed free-choice, commencing when the calves were ten days of age. Water and trace-mineralized salt were available free-choice throughout the experiment.

The calves were tied in individual stalls with expanded metal floors; no bedding was used. The calves were weighed at birth and every Wednesday thereafter. Each calf completed the experiment on the Wednesday closest to a 90-day period after birth.

Metabolism Studies

Six bull calves, two from each treatment, were used in metabolism trials. The trials were conducted for a period of 8 days, commencing when the calves were 72 to 76 days of age. In order to compare apparent digestion coefficients obtained by an indicator method with those obtained by total collection, 0.5 percent chromic oxide was mixed into each calf meal (Maynard and Loosli, 1969). This calf meal containing chromic oxide was fed for 8 days. After a 3-day preliminary period each calf was placed in a metabolism crate (Beacom and Thompson, 1963) for a 5-day collection period. Fecal grab samples were taken daily at 8 A.M., 11 A.M., 2 P.M. and 5 P.M., placed in polyethylene bags and stored at -3°C. At the end of the trial the 20 grab samples were placed together to make one sample for subsequent analyses.

In addition to the fecal grab samples, the total daily fecal excretion was collected once daily, the total weight was recorded and the feces was stored at -3°C in polyethylene bags. At the end of the trial the five samples were added together to make one sample for subsequent analysis.

Total urine excreted in the 5-day period was collected in a plastic bucket containing 15 ml of 50 percent (v/v) sulphuric acid as a preservative. The total volume was measured and 10 percent of the urine was placed in a polyethylene bottle and stored at 4°C.

Fecal samples were dried in a forced-draught oven at 70°C for 48 hours and ground in a laboratory mill. Dry matter and nitrogen in feed and fecal samples and nitrogen in urine samples were determined by AOAC (1965) methods. Dry matter in urine was determined by freeze-drying for 48 hours at -70°C and 30 microns of mercury pressure. Gross energy in feed, feces and dried urine was determined in a Parr oxygen bomb calorimeter.

Chromic oxide concentration in feed and fecal samples was determined by atomic absorption spectrophotometry as described by Williams et al. (1962).

Samples for Blood Urea Nitrogen Analyses

Blood samples were taken from each calf in the experiment at 80 to 84 days of age. Approximately 20 ml whole blood was drawn from the jugular vein into vacuum tubes containing 17 mg lithium oxalate. Each sample was centrifuged for 15 minutes at 1850 x g and the supernatant plasma was removed with a glass dropper and stored at -18°C in glass vials.

Blood urea nitrogen in plasma samples was determined by the method of Fawcett and Scott (1960).

Dissection Studies

At the end of the experiment, the six bull calves used in the metabolism studies were slaughtered. One half of each carcass was retained for studies of carcass composition as described by Berg and Mukhoty (1970). Each half-carcass was dissected to provide individual muscles, bones and tendons, and fat. The weights of total muscle, bones plus tendons, and total fat were recorded.

Statistical Analyses

Statistical analysis of the data involved the analysis of variance. The computer program for analysis of variance written by Smillie (1968) was used. Duncan's New Multiple Range Test (Steel and Torrie, 1960) was used to compare differences between means.

Results and Discussion

Average Feed Consumption

There were slight differences noted in milk consumption (Table 2). A few calves were unable to consume the maximum daily allowance of whole milk. Four calves (2 in Treatment I and 2 in Treatment II) did not consume sufficient calf meal at 28 days of age to permit weaning; these calves were fed additional milk or milk replacer for periods of 8 to 24 days until they were consuming enough calf meal and were vigorous enough to be weaned.

On the average, calves fed calf meal supplemented with soybean meal consumed the least calf meal up to 28 days of age (Table 2). The calf meal containing 0.75 percent urea appeared to be the most acceptable, since more of it was eaten than of the other calf meals. However, total consumption of calf meal was relatively small prior to weaning at 28 days of age, and most of it was consumed in the fourth week when the calves became hungry because of restriction in the amount of milk replacer offered.

After weaning, calves in Treatment I ate slightly more feed daily than those in Treatment II fed the calf meal with 0.5 percent urea (Table 2); they consumed 5.7 percent more calf meal daily during the experimental period of about 90 days. This small difference indicates that there was little difference in acceptability of the two calf meals. Calves in Treatment III consumed more calf meal daily throughout the experiment and averaged 19.8 percent more per day than the calves in Treatment I. This may be attributed to increased acceptability of this calf meal because of the higher level of dried molasses, or to increased

Table 2

Average daily feed consumption.

Treatment	I soybean meal	II 0.5% urea	III 0.75% urea	Standard error of the mean $S\bar{x}$
Average Pre-weaning Feed Intake (kg)				
Total whole milk	24.1	22.4	23.2	0.76
Total milk replacer	7.9	8.2	7.8	0.37
Total calf meal	5.4	6.5	8.2	0.81
Average Daily Calf Meal (kg)				
28 to 60 days	1.24	1.08	1.49	0.094
60 days to end of experiment	1.88	1.85	2.21	0.146
28 days to end of experiment	1.54	1.44	1.82	0.108
Birth to end of experiment	1.11	1.05	1.33	0.077

growth rate resulting from the higher level of nitrogen in the calf meal. Faster rate of growth would be associated with increased feed intake. The daily intake of calf meal by calves in Treatment III was significantly higher ($P < 0.05$) than for the other calves in the period 28 to 60 days of age. The other differences were not significant ($P < 0.05$) because of the large variability in feed consumption among calves in each treatment.

The data of feed intake obtained in this experiment do not agree with those reported elsewhere. Nelson et al. (1966) noted a reduction in feed intake when calves were fed calf meals containing 16.5 percent crude protein, with urea supplying 24 to 48 percent of the total nitrogen, as compared with a calf meal of approximately

equal nitrogen content supplemented with soybean meal. Miron et al. (1968) also obtained a reduction in feed intake (non-significant) when calves were fed a calf meal containing 18 percent crude protein, but containing 1.9 percent urea in place of 15 percent soybean meal. The high levels of urea used in those experiments might have caused the depression in feed intake, since urea is noted to be an unpalatable feed ingredient (Bartlett and Cotton, 1938). In the present experiment urea provided 9.6 percent of the total nitrogen in the ration fed to calves in Treatment II and 14.8 percent of the total nitrogen in the ration fed to calves in Treatment III.

On the average, bull calves consumed slightly more feed than heifer calves (1.24 versus 1.09 kg daily), but significant differences were not detected ($P < 0.05$).

Average Daily Gain

Average daily gain (Table 3) followed trends similar to those noted for feed consumption. Calves in Treatment I had slightly faster gains throughout the experiment than those in Treatment II, whereas the fastest gains were obtained by calves in Treatment III. The calves in Treatment III gained 28 percent faster than calves in Treatment II from birth to the end of the experiment. None of the differences were significant because of the variability within the treatments. Calves in all three treatments had similar gains exceeding 0.6 kg daily after 60 days of age. Presumably, the protein requirements would not be a limiting factor after the calves were two months of age. It is shown by the cumulative weight gains (Figure 2) that the major differences in weight gains occurred during

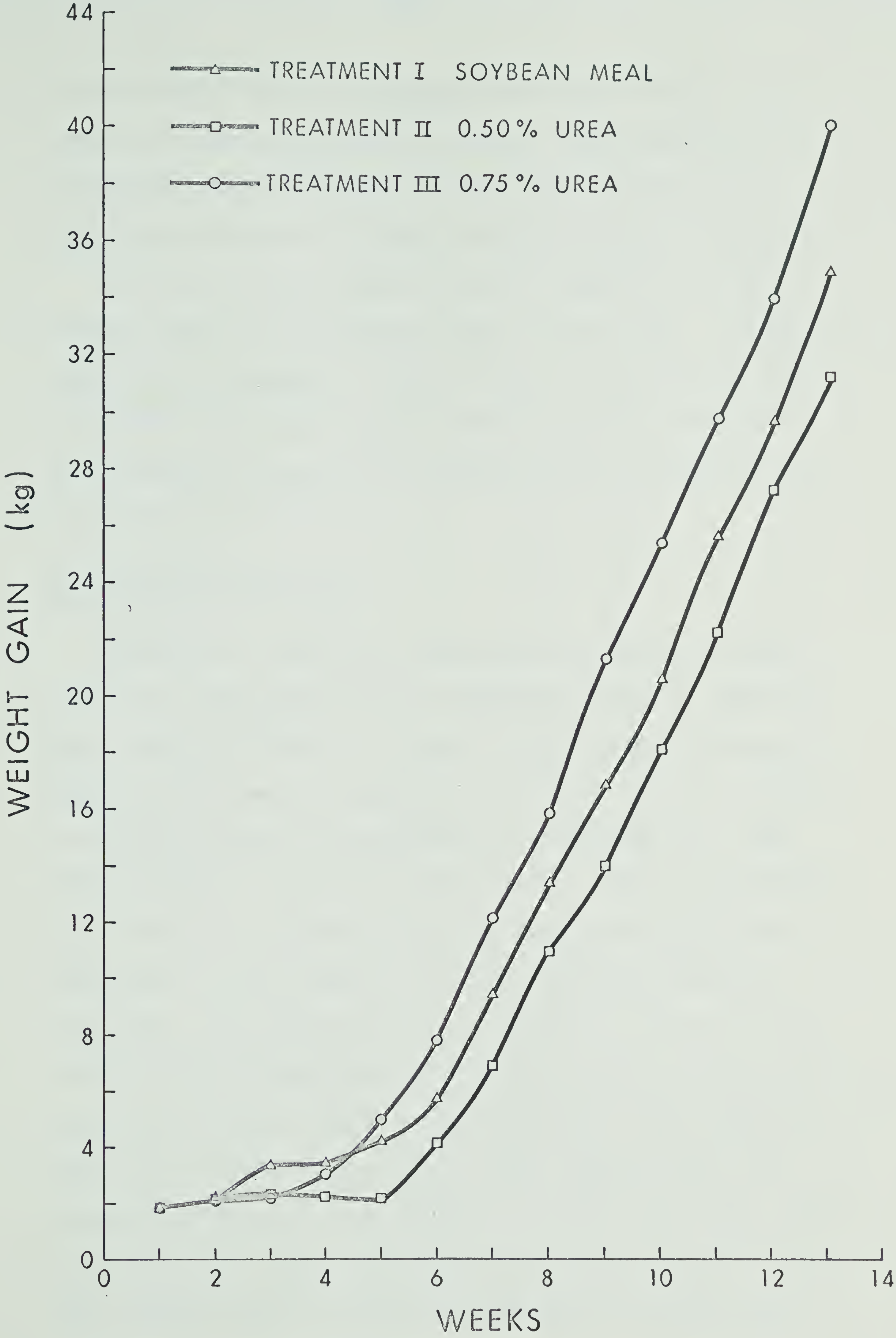

Figure 2. Cumulative weight gain.

the first six weeks and that average daily gains thereafter were similar for calves in all three treatments. This observation is in agreement with the results of Nelson (1970). There were no significant differences ($P < 0.05$) in daily gains between sexes.

The data of average daily gain do not suggest that urea-nitrogen proved to be a limiting factor in growth rate. The low feed intake in Treatment II calves appeared to be responsible for the slightly lower growth rate of these calves, but additional urea nitrogen in Treatment III was associated with increased feed consumption and growth rate.

Feed Utilization Efficiency

There were no significant differences ($P < 0.05$) in calf meal consumed per unit gain during the experiment (Table 3). Nevertheless, calves in Treatment I consumed the least feed per unit gain suggesting that the urea-supplemented calf meals were not used quite as efficiently as the one supplemented with soybean meal. This would be expected with calves in Treatment II. Since they ate less daily feed and gained less daily weight than calves in the other treatments, a higher proportion of their feed would be used for maintenance. The opposite would be expected with calves in Treatment III. They consumed the most feed and gained the fastest which should result in more efficient feed utilization. The results of Nelson *et al*. (1966) and Kay *et al*. (1967) indicated that urea supplementation of calf meal resulted in a lower feed intake, slower growth rate and a higher requirement of feed per unit gain. Presumably, this was caused by reduced acceptability of the calf meals

Table 3

Average weight gain and feed utilization.

Treatment	I soybean meal	II 0.5% urea	III 0.75% urea	Standard error of the mean $S\bar{x}$
Average final weight, kg	75.0	70.5	81.1	----
Average initial weight, kg	40.1	39.1	40.8	----
Average total gain, kg	34.9	31.4	40.2	----
Average Daily Gain (kg)				
28 to 60 days	0.40	0.36	0.56	0.053
60 days to end of experiment	0.66	0.62	0.68	0.078
28 days to end of experiment	0.52	0.48	0.62	0.047
Birth to end of experiment	0.40	0.36	0.46	0.036
Average kg Feed/kg Gain				
28 to 60 days	3.37	4.00	2.80	0.317
60 days to end of experiment	2.88	3.15	4.10	0.578
28 days to end of experiment	2.96	3.10	3.00	0.163
Birth to end of experiment	2.82	3.16	2.94	0.195

because of the high levels of urea used in those experiments.

Coefficients of Apparent Digestion

The use of chromic oxide did not provide reliable results in determining apparent digestibility of dry matter (Table 4). There was little variation between animals within treatments, but in two treatments (Treatment I and III) the results were much lower than those obtained by the conventional method of total collection of feces.

Low coefficients obtained with the indicator method would arise from incomplete recovery of the chromic oxide fed, resulting in overestimation of the total feces excreted. This may be attributed to chromic oxide sifting out of the ration, and fluctuations in the rate of excretion during the day. It has been noted in other reports (Clanton, 1962; Hardison *et al.*, 1959; Hattan and Owen, 1970; McGuire *et al.*, 1966; Milligan, 1967a) that chromic oxide resulted in low coefficients of apparent digestibility because of incomplete recovery of chromic oxide administered. Special precautions such as pelleting, (McGuire *et al.*, 1966) and cooking part of the ration with chromic oxide (Hattan and Owen, 1970) did not overcome the problem of low recovery of chromic oxide.

It seems apparent that chromic oxide is not a reliable indicator material when used in an all-concentrate diet fed to young ruminants. Consequently, the total fecal collection method was used to calculate coefficients of apparent digestibility.

Apparent digestibility of dry matter was significantly higher ($P < 0.05$) in the calf meal supplemented with soybean meal than in the calf meal supplemented with 0.50 percent urea. There were no significant differences ($P < 0.05$) in apparent digestibility of dry matter between the calf meals containing 0.75 percent urea and soybean meal. Apparent digestibility of gross energy was significantly higher ($P < 0.05$) in the calf meal supplemented with soybean meal than in those supplemented with urea (Table 4). These results are in agreement with those of Kay *et al.* (1967) and Stobo *et al.* (1967).

Although the apparent digestibility of gross energy was lower in the calf meal fed in Treatment III than in that fed in Treatment I,

calves in Treatment III consumed about 5.2 percent (261.08 kcal) more digestible energy daily than calves in Treatment I. This could account for the slightly faster growth rate of calves in Treatment III.

Apparent digestibility of nitrogen (Table 4) was significantly higher in the calf meal fed in Treatment I than in those fed in Treatment II ($P < 0.01$) and in Treatment III ($P < 0.05$). The apparent digestibility of nitrogen in the calf meal with 0.75 percent urea was significantly higher ($P < 0.05$) than in the one with 0.5 percent urea.

Nitrogen Retention

Calves in Treatment I retained an average of 14.4 grams of nitrogen daily which was 42.7 percent of the nitrogen consumed (Table 5). Calves in Treatment III consumed more nitrogen than those in Treatment I, had a higher rate of fecal excretion resulting in a lower apparent digestibility (Table 4), but retained 18.3 grams of nitrogen daily, which was 39.6 percent of the nitrogen consumed (Table 5). This was only slightly lower than the percentage retention obtained with calves in Treatment I. This suggests that nitrogen from urea was being used for protein synthesis by bacteria in the rumen, and that there was no marked increase in absorption of ammonia into the blood for subsequent elimination in the urine. There is a possibility that reduced digestibility of nitrogen in the calf meals containing urea might be due, at least in part, to rapid passage of some of the urea into the omasum before hydrolysis took place. With an all-concentrate ration, some of the ingested feed could pass directly through the reticulo-omasal orifice. Some of the urea might then be excreted directly in the feces.

Results in Treatment II were not as clear. Calf No. 932 had a high rate of fecal excretion of nitrogen (Table 5), resulting in

Table 4

Coefficients of apparent digestibility.

Treatment	I soybean meal			II 0.5% urea			III 0.75% urea			Standard error of the mean $S\bar{x}$
Calf No.	930	937	Ave.	932	938	Ave.	935	947	Ave.	
Dry matter, (chromic oxide) %	68.94	69.03	68.98	67.90	67.76	67.83	53.55	54.23	53.89	0.185
Dry matter, (total collection) %	75.33	80.26	77.80	65.70	69.12	67.41	67.53	70.83	69.18	1.977
Gross energy, %	74.74	79.74	77.24	65.49	68.12	66.80	67.68	69.76	68.72	1.738
Nitrogen, %	74.42	74.14	74.28	62.09	61.19	61.64	68.56	71.14	69.85	0.793

Table 5

Average daily nitrogen balance data.

Treatment	I soybean meal			II 0.5% urea			III 0.75% urea			Standard error of the mean
Calf No.	930	937	Ave.	932	938	Ave.	935	947	Ave.	$S\bar{x}$
N_2 consumed daily, g	36.3	31.5	33.9	37.4	24.1	30.8	39.0	54.3	46.7	6.02
N_2 in feces daily, g	9.3	8.2	8.7	14.2	9.4	11.8	12.3	15.7	14.0	1.74
N_2 in urine daily, g	12.7	9.0	10.8	7.4	13.7	10.5	10.3	18.5	14.4	3.18
N_2 retained daily, g	14.4	14.4	14.4	15.8	1.0	8.4	16.4	20.1	18.3	4.39
N_2 retention, %	39.6	45.8	42.7	42.2	4.3	23.2	42.1	37.0	39.6	11.8

low apparent digestibility (Table 4). However, urinary excretion by this calf was low, so that it retained slightly more nitrogen than calves in Treatment I and a similar percentage (Table 5). However, calf no. 938 had a high rate of urinary excretion, resulting in very low daily and percentage retention. This suggests that urea-nitrogen was not well utilized by microorganisms in the rumen of this calf. Ammonia from hydrolysis of the urea might have been absorbed into the blood and subsequently eliminated in the urine. However, it is not clear why this calf should have differed markedly from the other three calves fed urea. Low feed intake might suggest a digestive disturbance, but this was not indicated by the data of digestibility. If data for calf no. 938 were excluded, it would appear that there were no marked differences in percentage nitrogen retention by calves fed calf meals containing soybean meal or the two levels of urea.

True Digestibility, True Retention and Biological Value of Nitrogen

A portion of the total nitrogen excretion in ruminants as well as in other mammals is accounted for by losses due to endogenous and metabolic fecal nitrogen (Blaxter and Wood, 1952). These fractions have been utilized by the body even though they appear as excretions (Maynard and Loosli, 1969). In the present study the values of these two excretions were computed and used to derive the true nitrogen digestibilities, true nitrogen retentions and the biological values of nitrogen (Table 6). Knowledge of the true nitrogen digestibility together with the amount of nitrogen supplied by the grain part of the

ration could be used to compute the amount of urea nitrogen which may have been utilized by the calves.

Coefficients of true nitrogen digestibility and percentages of true nitrogen retention were similar to the trend observed in coefficients of apparent nitrogen digestibility and apparent nitrogen retention discussed earlier. Calves fed the soybean meal-supplemented ration (Treatment I) had higher values of true digestibility and true retention of nitrogen than calves fed the urea-supplemented rations (Treatments II and III). True digestibility of nitrogen was significantly higher for calves in Treatment I ($P < 0.01$) than for the calves in Treatments II and III. However, there were no significant differences ($P < 0.05$) among the treatments with regard to true retention of nitrogen.

There was no apparent difference in biological value of nitrogen in rations fed to calves in Treatments I and III (Table 6). This suggests that nitrogen from urea in Treatment III was utilized in protein synthesis in the rumen, otherwise the biological value would be lower than that obtained in Treatment I. Results with calf no. 932 in Treatment II suggested a high biological value of nitrogen, whereas a very low biological value was obtained with calf no. 938. It was apparent that calf no. 938 was not using nitrogen in the ration efficiently for protein synthesis.

The observations on true daily retention of nitrogen agree with the growth pattern noted earlier (Figure 2). Calves in Treatments I and II (except for calf no. 938) had similar nitrogen retentions and had similar growth rates after 6 weeks of age, whereas calves in Treatment III retained more nitrogen daily and grew slightly faster.

Table 6

True digestibility, retention and biological value of nitrogen.

Treatment	I soybean meal			II 0.50% urea			III 0.75% urea			Standard error of the mean
Calf No.	930	937	Ave.	932	938	Ave.	935	947	Ave.	$S\bar{x}$
Daily MFN[1], g	7.48	6.44	6.96	7.31	4.77	6.04	7.48	9.70	8.59	-----
Daily EUN[2], g	2.40	2.70	2.29	1.98	2.72	2.35	2.67	2.49	2.58	-----
True digestibility[3], %	95.02	94.55	94.78	81.31	80.79	81.06	87.73	89.01	88.37	0.446
True retention[4], g/day	24.26	23.04	23.65	25.19	8.53	16.86	26.61	32.34	29.48	5.006
True retention[5], %	66.78	73.05	69.92	67.26	32.36	49.81	68.18	59.55	63.68	9.714
Biological value[6], %	70.28	77.26	73.77	82.43	43.68	63.06	77.72	66.90	72.31	12.081

1 Daily metabolic fecal nitrogen was considered to be 2.811 g protein per 100 g dry matter ingested or 2.811/6.25 g nitrogen per 100 g dry matter ingested (Gardner, 1968).

2 Daily endogenous urinary nitrogen was calculated on the basis of 1.5 mg of endogenous nitrogen excretion per kcal of basal heat (BM = $70W^{0.75}$).

3 True nitrogen digestibility, % $= \dfrac{N_2 \text{ consumed} - \text{fecal } N_2 + \text{MFN}}{N_2 \text{ consumed}} \times 100$

4 True nitrogen retention, g/day $= N_2 \text{ consumed} - (\text{Fecal } N_2 - \text{MFN}) - (\text{Urinary } N_2 - \text{EUN})$

5 True nitrogen retention, % $= \dfrac{N_2 \text{ consumed} - (\text{Fecal } N_2 - \text{MFN}) - (\text{Urinary } N_2 - \text{EUN})}{N_2 \text{ consumed}} \times 100$

6 Biological value, % $= \dfrac{N_2 \text{ consumed} - (\text{Fecal } N_2 - \text{MFN}) - (\text{Urinary } N_2 - \text{EUN})}{N_2 \text{ consumed} - (\text{Fecal } N_2 - \text{MFN})} \times 100$

Blood Urea Concentrations

Calves fed the soybean meal-supplemented ration had a slightly lower average blood urea concentration than calves fed urea-supplemented rations (Table 7), but none of the differences were significant ($P < 0.05$). The values were similar to those obtained by Wells (1969). The non-significant increase in blood urea of calves fed the urea-supplemented calf meals suggests that supplemental urea did not result in excessive absorption of ammonia into the blood stream, and therefore ammonia released in the rumen may have been used in synthesis of protein. There were no significant differences ($P < 0.05$) in blood urea concentrations between sexes.

Table 7

Average blood urea concentration (mgm urea/100 ml plasma).

Treatment	I soybean meal	II 0.50% urea	III 0.75% urea	Standard error of the mean $S\bar{x}$
Male	14.16	17.84	15.60	----
Female	11.08	18.62	19.02	----
Average	12.62	18.26	17.32	2.020

It was expected that calf no. 938 (Treatment II) might have a high level of blood urea since results with this calf indicated low retention and low biological value of nitrogen. However, blood urea concentration for this calf was only 19.52 mgm/100 ml plasma. This suggests that it was affected by some metabolic disturbance during

the metabolism trial, and that it utilized nitrogen from urea as well as the other calves.

Carcass Composition

Calves fed rations supplemented with urea gained slightly more muscle per day and, therefore, percentage muscle in their carcasses at slaughter was slightly higher than that of calves fed the soybean meal-supplemented ration (Table 8). Calves in Treatment III had a slightly higher percentage of muscle than calves in Treatment II. However, these differences were not significant ($P < 0.05$).

The percentage of carcass fat was quite similar for all calves, with the urea treatments showing slightly higher average fat percentages than the soybean meal treatment. The lack of significance in carcass fat and muscle among the treatments suggests that all calves were generally lean and that muscle development proceeded at the same rate.

The skeletal development tended to be similar in every treatment (Table 8). The muscle to bone ratio was higher in calves fed urea-supplemented rations than in calves fed the soybean meal-supplemented ration. The muscle to fat ratio was higher for calves in Treatment I than for calves in Treatments II and III. None of these differences was significant ($P < 0.05$).

The carcass composition for all treatments appeared to be similar in pattern, with muscle comprising 64 to 66 percent, fat comprising about 9 to 10 percent and bone accounting for about 24 to 25 percent of carcass (Table 8). These results are in agreement with those of Moulton _et al_. (1922). The values obtained by these workers were from

Table 8

Carcass composition.

Treatment	I soybean meal			II 0.5% urea			III 0.75% urea				
Calf No.	930	937		932	938		935	947			
			Ave.			Ave.			Ave.	Standard error of the mean $S_{\bar{x}}$	Significance test
Age (days)	89	86	87.5	86	89	87.5	87	88	87.5	-----	
Liveweight (kg)	70.5	62.5	66.5	73.5	82.5	78.0	78.5	88.5	83.5	4.25	N.S.
Liveweight gain/day (kg)	0.32	0.28	0.30	0.43	0.38	0.40	0.48	0.56	0.52	0.028	*
Muscle per day (kg)	0.27	0.25	0.26	0.28	0.34	0.31	0.32	0.35	0.33	0.020	N.S.
Fat per day (kg)	0.04	0.04	0.04	0.05	0.05	0.05	0.06	0.04	0.05	0.000	N.S.
Percent muscle	66.5	61.6	64.1	62.5	66.2	64.4	65.8	66.9	66.3	1.80	N.S.
Percent bone (+ tendons)	24.4	29.3	26.8	26.3	24.6	25.4	22.4	24.6	23.5	0.80	N.S.
Percent fat	9.1	9.1	9.1	11.2	9.2	10.2	11.8	8.5	10.2	1.10	N.S.
Muscle to bone ratio	2.72	2.10	2.41	2.38	2.69	2.54	2.92	2.72	2.82	0.208	N.S.
Muscle to fat ratio	7.33	6.79	7.06	5.54	7.17	6.35	5.60	7.83	6.60	0.806	N.S.

* Significant at 5% level ($P < 0.05$).

the carcass of a normally fed steer of 3 months of age, with 25.527 percent bone, 66.551 percent lean and 6.535 percent fatty tissue. Butterfield _et al._ (1966) obtained similar values to those reported in this study with calves slaughtered at 12 weeks of age. However, fat percentage for calves used in their work was considerably lower than the values reported in the present study.

General Discussion

Higher averages in feed consumption and weight gain were obtained with calves fed calf meal containing 0.75 percent urea, than with calves fed rations containing 0.50 percent urea and soybean meal. Although calves fed the calf meal containing 0.50 percent urea had the lowest average feed intake and weight gain and consumed the most feed per unit weight gain, feed intake and weight gain by these calves was almost as high as that of calves fed the calf meal containing soybean meal.

These results suggest that there was no significant evidence of urea reducing the acceptability of the rations and, therefore, no adverse effect of urea on feed intake of calf meal as was reported by Kay et al. (1967). This lack of adverse effect might be attributed to the low levels of urea used in this experiment, as well as to the effect of dried molasses which should improve acceptability of a concentrate mixture.

Another possible explanation for the high feed intake and weight gain of calves fed the ration containing 0.75 percent urea would be the higher level of nitrogen contained in that ration. The high level of nitrogen was associated with increased nitrogen intake and retention and increased growth rate. The faster growth rate of these calves after weaning would increase their feed requirement as compared with calves in the other treatments.

Although the calf meal containing soybean meal was used more efficiently for weight gain than the rations containing urea, there was no marked advantage of the soybean meal ration over the ration containing 0.75 percent urea. Because of the lower apparent

digestibility of gross energy (Table 4), in calves fed the ration supplemented with 0.75 percent urea, calves on this ration used digestible energy more efficiently for weight gain than did the calves fed the soybean meal-supplemented ration.

The lower digestibility of dry matter and gross energy in rations containing urea cannot be explained. Differences in feed intake or rate of feed passage out of the rumen should not have had marked effects on digestibility. Therefore differences obtained may be attributed, at least in part, to variability associated with the small number of observations, as well as to actual differences between the rations. Wells (1969) found little difference in digestibility of dry matter and gross energy in a similar calf meal containing urea and a concentrate mixture containing soybean meal.

Calves fed the calf meals containing urea excreted more fecal nitrogen than those fed the soybean meal ration, resulting in lower digestibility of nitrogen in the rations containing urea.

Data of nitrogen retention and biological value suggest that three of the four calves used nitrogen in the urea rations as efficiently as calves fed the soybean meal ration. The absence of a marked increase in blood urea levels in calves fed the urea rations also suggests that nitrogen from urea was being used in protein synthesis, otherwise it would be expected that absorption of ammonia into the blood would result in high blood urea values. The fourth calf did not differ in digestibility of nitrogen from the other three, but had low retention of nitrogen resulting in low biological value. Nevertheless, it did not exhibit a high level of blood urea, suggesting that its performance during the metabolism trial could have been

affected by a metabolic disturbance, rather than by inability to utilize nitrogen from urea. In general, the data of growth rate, digestibility, nitrogen retention and biological value of nitrogen suggest that nitrogen from urea was used for growth or carcass development and had no deleterious effect when compared with soybean meal.

Further evidence that urea nitrogen was being used for growth by calves fed the ration supplemented with 0.75 percent urea can be obtained by calculating whether all nitrogen truly digested came from the grain portion of the ration or whether some of it came from urea. Such calculations can be done as follows:

Crude protein content of the ration (by analysis) = 14.39%

Therefore nitrogen content of the ration $= \frac{14.39}{6.25} = 2.30\%$

Crude protein supplied by urea in the ration = 2.11%

Therefore nitrogen from urea in the ration $= \frac{2.11}{6.25} = 0.34\%$

Percent of ration nitrogen supplied by urea $= \frac{0.34}{2.30} \times 100 = 14.78\%$

Average nitrogen consumption = 46.67 g/day (Table 5)

Urea nitrogen consumed $= \frac{14.78}{100} \times 46.67 = 6.90$ g/day

Therefore nitrogen from grain in the ration = 46.67 - 6.90 = 39.77 g/day

Fecal nitrogen = 13.97 g/day (Table 5)

Metabolic fecal nitrogen (MFN) = 8.59 g/day (Table 6)

Nitrogen truly digested = N_2 consumed - (Fecal N_2 - MFN)

= 46.67 - (13.97 - 8.59) = 41.29 g/day

Therefore possible nitrogen supplied by urea = 41.29 - 39.77

= 1.52 g/day

As the grain portion of the ration, by calculation, could supply no more than 39.77 grams per day of the calves' average nitrogen intake, at least 1.52 grams per day of nitrogen truly digested was urea nitrogen. However, not all of the grain nitrogen would be digested and therefore urea nitrogen would comprise more of the truly digested nitrogen than implied in the conclusions above.

Calculated results for the true nitrogen digestibility, retention and biological value indicate that the nitrogen from the soybean meal-supplemented calf meal was utilized slightly better than nitrogen from the calf meal supplemented with 0.75 percent urea. Since the biological values for the two calf meals were almost the same, it can be concluded that urea nitrogen was used for growth as efficiently as soybean meal nitrogen.

Carcass composition data indicated that muscle development for calves in all treatments was similar. When slaughtered, at about 90 days of age, calves fed urea-supplemented rations had slightly more muscle than calves fed the soybean meal-supplemented ration. The fat percentage was also slightly higher for the calves fed urea-supplemented calf meals than for the calves fed soybean meal-supplemented ration. However, calves fed soybean meal-supplemented ration had slightly heavier bone than calves fed the urea-supplemented rations. The carcass composition data for all treatments agree with the results of Butterfield et al. (1966), Moulton et al. (1922) and Mukhoty et al. (1970).

In general, observations in these experiments suggest that urea is a good source of supplemental nitrogen. Calves can be early-weaned to urea-containing high-energy concentrate calf meals.

Satisfactory feed intake and weight gain observed among the urea-fed calves suggest that the urea-supplemented calf meals were well accepted and utilized by the calves. High nitrogen retention levels and normal carcass development of these calves supply further evidence that non-protein nitrogen is of significance even in young ruminants.

Summary and Conclusions

Calves fed the calf meal supplemented with 0.75 percent urea consumed more average feed, had higher average weight gain, retained more average nitrogen, and gained more average carcass muscle per day than calves fed the calf meal supplemented with either soybean meal or 0.50 percent urea. However, calves fed the calf meal supplemented with soybean meal digested dry matter, gross energy and nitrogen better than calves fed calf meals supplemented with urea.

Efficiency of feed utilization was better for calves fed the soybean meal-supplemented calf meal than for calves fed the urea-supplemented rations. However, calves fed the urea-supplemented rations utilized digestible energy in the rations more efficiently than calves fed the soybean meal-supplemented ration.

Average concentrations of blood urea nitrogen were approximately the same for all calves. This suggested that urea nitrogen in calves fed urea-supplemented calf meals was not converted into urea in excessive amounts by the liver and returned in the bloodstream to be excreted in urine.

On the basis of the results obtained in this study, it is suggested that urea is a useful source of supplemental nitrogen at low levels in high-concentrate calf meals, when fed to early-weaned calves. Non-protein nitrogen from urea in this experiment appeared to be used by rumen bacteria for microbial protein synthesis with an efficiency comparable to that of soybean meal nitrogen.

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Appendix

Sums of squares and mean squares obtained by analysis of variance

Variable	Source of variation	Degrees of freedom	Sums of squares	Mean squares
Average daily calf meal 28 to 60 days	Ration	2	0.70	0.35*
	Sex	1	0.08	0.09
	Ration X Sex	2	0.02	0.01
	Error	18	1.28	0.07
	Total	23	2.09	
60 days to end of experiment	Ration	2	0.64	0.32
	Sex	1	0.60	0.60
	Ration X Sex	2	0.15	0.08
	Error	18	3.05	0.17
	Total	23	4.44	
28 days to end of experiment	Ration	2	0.63	0.32
	Sex	1	0.28	0.28
	Ration X Sex	2	0.04	0.02
	Error	18	1.69	0.09
	Total	23	2.64	
Birth to end of experiment	Ration	2	0.35	0.17
	Sex	1	0.15	0.15
	Ration X Sex	2	0.02	0.01
	Error	18	0.86	0.05
	Total	23	1.38	
Average daily gain 28 to 60 days	Ration	2	0.17	0.09
	Sex	1	0.04	0.04
	Ration X Sex	2	0.03	0.01
	Error	18	0.41	0.02
	Total	23	0.64	

Variable	Source of variation	Degrees of freedom	Sums of squares	Mean squares
60 days to end of experiment	Ration	2	0.01	0.01
	Sex	1	0.00	0.00
	Ration X Sex	2	0.03	0.02
	Error	18	0.89	0.05
	Total	23	0.93	
28 days to end of experiment	Ration	2	0.06	0.03
	Sex	1	0.00	0.00
	Ration X Sex	2	0.00	0.00
	Error	18	0.33	0.02
	Total	23	0.39	
Birth to end of experiment	Ration	2	0.04	0.02
	Sex	1	0.01	0.01
	Ration X Sex	2	0.00	0.00
	Error	18	0.20	0.01
	Total	23	0.25	
Average feed/kg gain 28 to 60 days	Ration	2	1.63	0.82
	Sex	1	0.13	0.13
	Ration X Sex	2	1.33	0.67
	Error	18	25.05	1.39
	Total	23	28.14	
60 days to end of experiment	Ration	2	6.55	4.27
	Sex	1	5.91	5.91
	Ration X Sex	2	8.35	4.17
	Error	18	48.13	2.67
	Total	23	68.93	

Variable	Source of variation	Degrees of freedom	Sums of squares	Mean squares
28 days to end of experiment	Ration	2	0.01	0.00
	Sex	1	0.32	0.32
	Ration X Sex	2	0.04	0.02
	Error	18	3.84	0.21
	Total	23	4.20	
Birth to end of experiment	Ration	2	0.50	0.25
	Sex	1	0.02	0.02
	Ration X Sex	2	0.09	0.04
	Error	18	5.48	0.30
	Total	23	6.10	
Coefficients of apparent digestibility				
Dry matter	Ration	2	123.47	61.73*
	Error	3	23.44	7.81
	Total	5	146.91	
Gross energy	Ration	2	123.43	61.72*
	Error	3	18.12	6.04
	Total	5	141.55	
Nitrogen	Ration	2	164.53	82.27**
	Error	3	3.77	1.26
	Total	5	168.30	
Nitrogen balance				
N_2 consumed daily	Ration	2	282.96	141.48
	Error	3	217.06	72.35
	Total	5	500.01	
N_2 in feces daily	Ration	2	27.76	13.88
	Error	3	18.13	6.04
	Total	5	45.89	

Variable	Source of variation	Degrees of freedom	Sums of squares	Mean squares
N_2 in urine daily	Ration	2	19.05	9.52
	Error	3	60.59	20.19
	Total	5	79.64	
Nitrogen retained daily	Ration	2	98.61	49.31
	Error	3	115.66	38.56
	Total	5	214.28	
Nitrogen retention percent	Ration	2	435.27	217.64
	Error	3	749.55	249.85
	Total	5	1184.82	
Blood urea	Ration	2	10.19	5.10
	Sex	1	0.18	0.18
	Ration X Sex	2	20.80	10.40
	Error	18	588.75	32.71
	Total	23	619.92	
Carcass composition percent muscle	Ration	2	5.99	2.99
	Error	3	19.40	6.47
	Total	5	25.40	
Percent bone	Ration	2	3.67	1.84
	Error	3	3.86	1.29
	Total	5	7.53	
Percent fat	Ration	2	1.69	0.85
	Error	3	7.23	2.41
	Total	5	8.92	
Muscle to bone ratio	Ration	2	0.18	0.09
	Error	3	0.26	0.09
	Total	5	0.44	

* $P < 0.05$.
** $P < 0.01$.